Thermoplastic Polyurethanes for the Manufacturing of Highly Dosed Oral Sustained Release Matrices via Hot Melt Extrusion and Injection Molding

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Abstract

This study evaluated thermoplastic polyurethanes (TPUR) as matrix excipients for the production of oral solid dosage forms via hot melt extrusion (HME) in combination with injection molding (IM). We demonstrated that TPURs enable the production of solid dispersions – crystalline API in a crystalline carrier – at an extrusion temperature below the drug melting temperature (Tm) with a drug content up to 65% (wt.%). The release of metoprolol tartrate was controlled over 24h, whereas a complete release of diprophylline was only possible in combination with a drug release modifier: polyethylene glycol 4000 (PEG 4000) or Tween 80. No burst release nor a change in tablet size and geometry was detected for any of the formulations after dissolution testing. The total matrix porosity increased gradually upon drug release. Oral administration of TPUR did not affect the GI ecosystem (pH, bacterial count, short chain fatty acids), monitored via the Simulator of the Human Intestinal Microbial Ecosystem (SHIME). The high drug load (65wt.%) in combination with (in-vitro and in-vivo) controlled release capacity of the formulations, is noteworthy in the field of formulations produced via HME/IM.

Introduction

Sustaining drug release from a dosage form after oral administration offers distinct advantages to chronic and poly-medicated patients: maintaining drug levels in the therapeutic range, lower dosing frequency, less side effects; resulting in a better patient compliance. While sustained drug release can be achieved based on the design of the dosage form (reservoir systems vs. matrices) and/or the physicochemical properties of the polymeric materials incorporated in the formulation (diffusion controlled vs. delayed polymer dissolution), hot melt extrusion (HME) (possibly in combination with injection molding) has been evaluated to manufacture sustained-release matrices using various polymers: ethylcellulose (EC) [1-3], hydroxypropyl (methyl) cellulose (HP(M)C) [4], ethylene vinyl acetate (EVA) [5-6], polyvinyl acetate (PVA) [7], poly lactic (co-glycolic) acid (PL(G)A) [8-9], silicone [10], polycaprolactone (PCL) [11-12], polyoxazolines [13], polyanhydrides [14], methacrylate copolymers (Eudragit® RS/RL) [15-16], and several lipid materials [17-19]. While sustained-release dosage forms have been successfully developed via HME using these polymers, a common drawback is that the drug load in these formulations is often low, either linked to processing issues during HME of formulations with a high drug load, or due to a significant burst release when less polymeric matrix former is incorporated in the formulation. Quinten et. al. [20], for instance, described that drug load in an acrylic polymermatrix was limited to 30% when processed via HME/IM, drug release from these matrices occurred in a first order manner via a combination of swelling and diffusion. Reitz et al. managed [21] to produce extrudates with 65wt.% diprophylline via solid lipid extrusion (with glycerol trimyristate as carrier) and identified the importance of drug particle size: although a larger drug particle size reduced the release rate and minimized the burst release, sustained release from these high dosed lipid matrices was limited (70% drug release after 1 and 3 h for small and large drug particle sizes, respectively). Another study [22] incorporated 30wt.%

diprophylline in Eudragit® S100 and Eudragit® L matrices, processed via HME at 160°C, and due to the complete dissolution of diprophylline in the matrix poor sustained release properties (>50% released within 2h in 1N HCl) were registered. It is well documented that a higher drug load enhanced the release rate from the matrix since more pores are created in the micro-capillary network of the insoluble matrix upon drug release, often combined with a burst release. To this end, the design of novel sustained release dosage forms using innovative polymeric materials with improved characteristics for controlled drug release is continuously under investigation.

Thermoplastic polyurethanes (TPUR) are inert, non-ionic, water-insoluble polymers that have been successfully used for many years as drug release controlling polymers in vaginal rings [23-25], stents [26], coatings [27] and implants [28]. Another important application of TPUR is in medical tubing as its superior mechanical properties (compared to polyvinyl chloride) allow the use of thinner walled tubes, even without a plasticizer [29]. The chemical structure of TPUR consists of alternating hard segments (HS) in a continuous phase of soft segments (SS) (Figure 1). Typically the SS phase is composed of a relatively long, flexible polyester or polyethers with a molecular weight of 1000-10.000g/mol. The HS can be composed of linear 4,4'-methylene diphenyl diisocyanate (MDI) or its hydrogenated form (HMDI), possibly linked to one another via a chain extender (often short chain diols). Thermodynamic incompatibility of both segments results to a certain degree of insolubility between HS and SS, yielding a microphase-separated (segmented) polymer. This generates a polymer with unique physicochemical characteristics: an elastomeric material with superior tensile strength, abrasion resistance, crack resistance, inherent lubricity and biocompatibility [30]. To this end, the HS contributes to the polymers' strength due to the formation of intermolecular hydrogen bonds between the urethane structures (NH-(C=O)-O) in each HS, while the SS fraction with a low

glass transition temperature (Tg) provides the polymers' elasticity [31]. TPUR are available in various molecular weights, different types (polyester, polyether) of SS, different SS lengths, and variable SS/HS ratios which makes them attractive candidates to alter drug release performances based on polymer composition.

In this study, we evaluated thermoplastic polyurethanes (TPUR) as matrix excipients for oral sustained release purposes. The formulations were produced via hot-melt extrusion followed by injection molding and were consequently (physicochemically) characterized. Metoprolol tartrate (MPT), theophylline (Th) and diprophylline (Dyph) were embedded as model drugs in the TPUR matrices.

In addition to the processing of TPUR via hot-melt extrusion and its physicochemical evaluation, an assessment of the safety of TPUR upon oral intake was performed via the SHIME (Simulator Human Intestinal Microbial Ecosystem) study. While this approach does not provide an entire toxicological report, it can give a first impression about the impact of the gastro-intestinal fluids on the integrity of TPUR and the effect of TPUR on the microbial flora. Upon oral intake, the formulation passes the intestinal environment where chemical and/or enzymatic reactions can affect the polymer structure. Also, the human colon, which is colonized by a complex microbial community, can modify the polymer structure and may in turn be influenced by exposure to the polymer.

Experimental Section

Materials

Various grades of TPUR (Table 1) with varying composition of the hard and soft segments and with variable SS/HS ratio (Table 1) were obtained from Merquinsa (a Lubrizol company, Ohio, USA): the Pearlbond polyurethanes (P520, P522, P523, P539) were non-medical grades, while the Tecoflex types (T85A, T100A, T72D) were medical grades (Figure 2). Metoprolol tartrate (MPT) (Esteve Quimica, Barcelona, Spain), theophylline (Th) and diprophylline (Dyph, 7-(2,3-dihydroxypropyl)-theophylline) (Sigma Aldrich, Bornem, Belgium) are embedded as model drugs in the TPUR matrices. Polyethylene glycol 4000 and Tween 80 were obtained from Fagron (Waregem, Belgium).

Size exclusion chromatography (SEC)

Size exclusion chromatography (SEC) was performed on a Waters instrument, equipped with 3 serial Polymer Standards Services columns (1 x GRAM Analytical 30 Å and 2 x GRAM Analytical 1000 Å, 10 µm particle size) at 35°C. Poly(methylmethacrylate) (PMMA) standards were used for calibration and dimethylacetamide (DMA), containing LiBr (0.42 g/mL) to increase polymer solubility, was used as solvent at a flow rate of 1 mL/min. 100 µL of a 10 mg/mL TPUR solution was injected onto the column. TPUR in the eluent was detected using a Waters 2414 refractive index detector. Molecular weights were determined using the Empower software (Waters, Zellik, Belgium).

Nuclear magnetic resonance (NMR)

NMR spectra were recorded on a Bruker AVANCE 300 spectrometer, using deuterated dimethylformamide (DMF) as solvent, to determine the polymer structure and SS/HS ratio. The spectra were analyzed with the ACD/Spec Manager software from ACD/Labs. ^{1}H-NMR (300 MHz, DMF-d7) of Pearlbond polymers: δ (ppm) = 1.37 (m, C*H*$_2$-CH$_2$-CH$_2$-O), 1.61 (m, CH$_2$-C*H*$_2$-CH$_2$-O), 2.33 (t, CH$_2$-CH$_2$-C*H*$_2$-(CO)), 3.86 (s, Ph-C*H*$_2$-Ph), 4.05 (t, CH$_2$-CH$_2$-C*H*$_2$-O), 7.17 (d, aromatic), 7.48 (d, aromatic), 9.45 (s, O(CO)N*H*). ^{1}H-NMR (300 MHz, DMF-d7) of Tecoflex polymers: δ (ppm) = 0.86 – 1.88 (br, C*H*$_2$ cyclic and C*H*$_2$-CH$_2$-O(CO)NH), 1.57 (br, C*H*$_2$ backbone pTHF), 3.39 (br, C*H*$_2$-O), 3.62 (br, O(CO)NH-C*H*), 3.98 (br, CH$_2$-C*H*$_2$-O(CO)NH), 6.71-7.03 (br, O(CO)N*H*).

Fourier-transform infrared spectroscopy

Attenuated total reflection Fourier-transform infrared (ATR FT-IR) spectroscopy was performed on the polymers before and after the SHIME-experiment in order to identify molecular changes. Spectra were recorded using a Nicolet iS5 ATR FT-IR spectrometer (Thermo Fisher Scientific). A diamond ATR crystal was pressed against the samples. Each spectrum was collected in the 4000 - 550 cm^{-1} range with a resolution of 2 cm^{-1} and averaged over 32 scans.

Thermal analysis

Thermogravimetric analysis (TGA 2950, TA instruments, Leatherhead, UK) was used to investigate the thermal stability of the polymers. The samples were equilibrated at 30°C and heated (10°C/min) to 500°C under an N_2 atmosphere.

The glass transition temperature (Tg) and melting point (T_m) of pure components, physical mixtures and injection molded tablets were analyzed in Tzero pans (TA instruments, Zellik, Belgium) by modulated differential scanning calorimetry (MDSC Q2000, TA Instruments, Leatherhead, UK) using a heating rate of 2°C/min. The modulation period and amplitude were set at 1min and 0.318°C, respectively (heat-iso method). Dry nitrogen at a flow rate of 50mL/min was used to purge the MDSC cell. Analysis of the thermal characteristics ($T_{m,}$ and T_g) was done via a heating/cool/heat run between -70°C and 75°C and between -70°C and 140°C for the physical mixtures with Pearlbond and Tecoflex polymers, respectively. The melting enthalpy (in the total heat flow signal), Tmelt-max (i.e. inflection point of melting endotherm) and Tmelt-onset (i.e. start of melting endotherm) were analyzed in the first heating cycle. The crystallinity was determined by dividing the API melting enthalpy in the formulation by the melting enthalpy of pure API components (=100% crystalline). Analysis of the glass transition temperature was done in the first and second heating cycle for injection molded tablets and physical mixtures, respectively. All results were analyzed using the TA Instruments Universal Analysis 2000 software.

Production of injection molded tablets

Physical mixtures, homogenized using mortar and pestle, of drug/polymer at a ratio of 50/50, 65/35 and 75/25 were extruded at 70°C for MPT-containing formulations, and at 140°C for Th- and Dyph-containing formulations using a lab-scale co-rotating twin-screw extruder at 100rpm (Haake MiniLab II Micro Compounder, Thermo Electron, Karlsruhe, Germany). The mixtures were manually fed into the extruder in order to avoid segregation due to differences in particle size between API (µm range) and TPUR (mm range). Immediately after HME, the thermoplastic melt was processed into biconvex tablets (diameter: 10mm/height: 5mm) via

injection molding (Haake MiniJet System, Thermo Electron). The injection pressure was 800bar during 10s, in combination with a post-pressure of 400bar for 5s. The temperature during injection molding was the same as during HME: 70 and 140°C for MPT-containing and Th- and Dyph-containing formulations, respectively.

Raman mapping

The homogeneity of the distribution of MPT in the tablets was evaluated by Raman microscopic mapping using a Raman Rxn1 Microprobe (Kaiser Optical Systems, Ann Arbor, MI, USA) equipped with an air-cooled CCD detector. The laser wavelength employed was a 785 nm from a Invictus NIR diode laser. The tablet surface was scanned by a 10x long working distance objective lens (spot size 50 μm) in area mapping mode using an exposure time of 4s and a step size of 50 μm in both the x (18 points) and y (13 points) direction (=234 spectra or 850 x 600μm per mapping segment). Six areas were analyzed in total. Data collection and data transfer were automated using HoloGRAMS™ data collection software (version 2.3.5, Kaiser Optical Systems), the HoloMAP™ data analysis software (version 2.3.5, Kaiser Optical Systems) and Matlab® software (version 7.1, The MathWorks, Natick, MA, USA). All spectra were reduced to 800-1500cm^{-1}, a spectral range which contains the fingerprint region of both components. The spectra were baseline corrected using Pearson's method and then normalized.

The 234 Raman spectra collected per monitored area were each introduced into a data matrix (**D**), resulting in a Raman data matrix per area. Each **D** was analyzed using multivariate curve resolution (MCR). MCR aims to obtain a clear description of the contribution of each pure component in the area from the overall measured variation in **D**. Hence, all collected spectra in the area are considered as the result of the additive contribution of all pure components involved

in the area. Therefore, MCR decomposes **D** into the contributions linked to each of the pure components in the system:

$$\mathbf{D} = \mathbf{CS} + \mathbf{E} \qquad \text{(equation 1)}$$

where **C** and **S** represent the concentration profiles and spectra, respectively. **E** is the error matrix, which is the residual variation of the dataset that is not related to any chemical contribution. Next, the working procedure of the resolution method started with the initial estimation of **C** and **S** and continued by optimizing iteratively the concentration and response profiles using the available information about the system. The introduction of this information was carried out through the implementation of constraints. Constraints are mathematical or chemical properties systematically fulfilled by the whole system or by some of its pure contributions. The constraint used for this study was the default assumption of non-negativity; that is, the data were decomposed as non-negative concentration times non-negative spectra [32].

Melt rheology

Melt rheology of TPUR was determined using an Anton Paar MCR301 (Oregon, USA) rheometer. The gap between plate spindle (diameter 25 mm) and plate was 1 mm. The strain amplitude and the angular frequency used were 1% and 10 rad/s, respectively. The Pearlbond and Tecoflex viscosities were measured at a temperature of 70 and 140°C, respectively.

Scanning electronic microscopy

IM tablets were sputtered with platinum using the JEOL JFC 1300 Auto Fine Coater (Jeol, Zaventem, Belgium). The samples were examined with a JEOL JSM 5600 LV scanning electron microscope (Jeol) at a magnification of 1000x.

He pycnometry

The tablet porosity was calculated based on the difference between the bulk and skeletal volume of the injection molded tablets. The skeletal volume of the tablets was measured (at different time points during dissolution experiments: 2, 4, 6, 8 and 24 h) via He pycnometry (AccuPyc 1330, Micromeritics, Norcross, USA). Prior to He pycnometry the tablets were dried for 2 days at 30°C. As no shrinkage or swelling of the injection molded tablets was observed immediately after injection molding nor after 24 h dissolution testing (verified by measuring tablet diameter and height using a digital slide caliper), the bulk volume of the tablets was determined from the dimensions of the mold. The tablet porosity (□) was calculated based on the following equation:

□ = [(bulk volume – skeletal volume)/bulk volume] x 100

(equation 2)

***In vitro* drug release**

Drug release from the injection molded tablets was determined using the paddle method on a VK 7010 dissolution system (VanKel Industries, New Jersey, USA) with a paddle speed of 100rpm. Distilled water was used as dissolution medium (900mL) at 37 ± 0.5°C. Samples were withdrawn at 0.5, 1, 2, 4, 6, 8, 12, 16, 20 and 24 h and spectrophotometrically analyzed for API concentration at 272nm for Th and 274nm for MPT and Dyph, respectively.

***In vivo* evaluation**

All procedures were performed in accordance with the guidelines and after approval by the Ethics Committee of the Faculty of Veterinary Medicine (Ghent University). To study the

influence of MPT concentration, 2 formulations were administrated to 6 dogs: (a) formulation F1: IM tablets containing 65 % MPT and 35 % Pearlbond 539 (equivalent to 239mg MPT), (b) formulation F2 (reference): Slow-Lopresor® 200 Divitabs® (Sankyo, Louvain-la-Neuve, Belgium), a commercial sustained release formulation consisting of matrix tablets containing 200 mg MPT.

All formulations were administrated to 6 male mixed-breed dogs (10 – 13 kg) in a cross-over study with a wash-out period of at least 8 days. Since the size of the molded tablets was fixed, different MPT doses (239 and 200 mg for F1 and F2, respectively) were administered. The pharmacokinetic profiles were normalized as linear pharmacokinetics have been reported for MPT in a dose range between 50 and 400 mg [33]. The dogs were fasted 12 h prior to administration and 12 h after administration, although water was available ad libitum. Before administration, an intravenous cannula was placed in the lateral saphenous and a blank blood sample was collected. The formulations were administrated with 20 mL water, and blood samples were collected in dry heparinized tubes at 0.5, 1, 2, 4, 6, 8, 12, 16, 20 and 24 h after administration. The obtained blood samples were centrifuged at 1500g during 5 min. A validated HPLC method [34] with fluorescence detection was used for the determination of MPT in dog plasma. The peak plasma concentration (C_{max}) and the time needed to reach the highest plasma concentration (t_{max}) were determined. The controlled release characteristics of the formulations were evaluated by means of the $HVD_{t50\%Cmax}$ defined by the period during which the plasma concentration exceeds 50 % of C_{max} [35-36]. The intact tablets, collected in the faeces of the dogs, were analyzed for their remaining MPT concentration. These tablets were crushed using mortar and pestle, suspended in 100mL demi water for 24 h and the MPT concentration in the supernatant was spectrophotometrically analyzed at 274nm.

Simulator of the Human Intestinal Microbial Ecosystem (SHIME)

The stability of TPUR grades T85A and P523 upon oral ingestion was evaluated in the Simulator of the Human Intestinal Microbial Ecosystem (SHIME), developed by the Laboratory of Microbial Ecology and Technology, Ghent University, as described earlier [37]. In brief, both polymers (2 g/l) were added to a standardized nutritional medium and incubated under simulated stomach conditions for 90 min (37°C, aerophilic conditions). Next, an appropriate amount of bile salts and digestive enzymes was added to simulate small intestinal conditions, and the samples were further incubated for 150 min (37°C, microaerophilic conditions). Finally, a complex microbial community was taken from the ascending colon compartment of the SHIME and added to the setup. The samples were further incubated for a period of 48 h (37°C, anaerobic conditions). All experiments were performed in triplicate. As a control, the same experiment was performed in parallel, without addition of TPUR. Both polymers were isolated from the stomach, small intestinal and colon incubation medium, and the polymer's integrity and structure was analyzed by means of MDSC and FTIR. The potential breakdown of the polymer by the intestinal microflora was indirectly assessed. Changes in composition or activity of the microbial community in the test with TPUR as compared to the control were used as marker for interaction between TPUR and the intestinal microbiota. Changes in composition were assessed by selective plate counting for different bacterial groups, as described by Possemiers et al. [38]. Effects on microbial activity were evaluated by pH measurements and by quantification of the concentrations of shortchain fatty acids (SCFA) in the samples at the beginning and end of the colonic incubations, as described earlier [38].

Results and Discussion

Processability via extrusion and injection molding

Initially, the maximum drug load of the formulations that allowed processing via HME and IM was determined. Using Pearlbond and Tecoflex polymers as matrix formers, two distinct factors were negatively influencing the production of high drug load matrices: processing temperature and powder fraction in the formulation. If the processing temperature exceeded the API's melting point (T_m), HME processing became impossible at high drug loads as the matrix former was not able to absorb the large amount of molten API, resulting in a too liquid phase without the plasticity required for the HME process. In addition, a large powder fraction in a formulation also compromised HME processing below the API's T_m as the high percentage of crystalline API resulted in a too high torque during HME. Therefore, the TPUR fraction in the formulation must be sufficient to provide sufficient plasticity during thermal processing.

Thermal processing of Pearlbond polymers was possible at a temperature of 70°C. Polycaprolactone (PCL, $(C_6H_{10}O_2)_n$), the semi-crystalline polyester soft segment (SS) in Pearlbond, has a Tg of around -60°C and melts at 55°C [39]. The combination of hard segments (HS), intermolecular connected via H-bonds, and the molten SS provides the polymer with sufficient plasticity needed for the extrusion process. This low processing temperature enabled the production of high drug loaded formulations with all three API's: MPT, Dyph and Th (T_m of 120, 160 and 270°C, respectively). The maximum drug load was 65wt.% API using P523 and P539 as matrix formers, while only 50wt.% API could be combined with P520 and P522. This difference in processability was correlated with the lower SS/HS ratios of P520 and P522 (Table 1). A higher fraction of hard segment (HS), methylene diphenyl diisocyanate (MDI) in Pearlbond, hampers the movement of SS, which makes the polymer more rigid and, hence, more

difficult to process. Thermal analysis confirmed this increase in rigidity as the change in heat capacity (ΔCp) at the Tg of P539, P523 and P520 was inversely correlated to the SS/HS ratio (Figure 3): a higher fraction of HS in TPUR increased the energy needed to transform the polymer from its glass state to a rubbery phase, reducing the processability of P520. These findings were also confirmed by melt rheology experiments of the polymers, as higher melt viscosities were measured at 70°C for P520 and P522, respectively (Table 1). The polymer melt viscosity could be correlated with their extrusion processability as the API did not dissolve in the polymer melt during HME/IM processing: no loss of API crystallinity was detected in the IM tablets based on the melting enthalpy of an MPT/P539 formulation containing 50 and 65% drug, corresponding to 99 and 100% MPT crystallinity, respectively. Similar results were obtained when the other Pearlbond/drug combinations were processed (data not shown).

The formulations based on the medical grade polyether TPUR (Tecoflex) could only be processed via extrusion and injection molding at a temperature of 140°C. This higher energy input, required for the Tecoflex polymers to provide sufficient plasticity to the formulation, is most likely linked to their chemical structure consisting of hydrogenated MDI (HMDI) as HS with a shorter and thus more rigid poly-tetrahydrofuran (pTHF, $(C_4H_8O)_n$) as SS [40]. They also contain a chain extender, most likely a butanediol, which increases the HS length, a significantly lower SS/HS ratio (Table 1) and a higher polymer melt viscosity. The higher processing temperature, however, excluded MPT from Tecoflex-based formulations as its melting point (T_m: 120°C) was below the process temperature, yielding a liquid mixture without the (thermo)plasticity required for HME. Using T85A and T100A as matrix former, formulations were produced up to an API content of 65wt.%, while even a drug load of 75wt.% was possible in combination with T72D. Processing of Dyph/Tecoflex mixtures at high drug load via HME

and IM was facilitated by the partial loss of Dyph crystallinity during HME: at a drug load of 65 wt%. in T72D and T100A matrices 63 and 70%, respectively, of crystalline Dyph was recovered in the IM tablets.

An additional advantage of TPUR is its inherent lubricity [30]. Previous studies considering the injection molding technique used a silicon-based anti-sticking spray to facilitate the release of the solidified tablets from the mold [2, 41] or had problems with the brittleness of their formulations [13, 42]. Using TPUR as matrix former, no sticking to the mold was observed, nor brittleness of the matrix after cooling.

In-vitro drug release

The influence of drug load on the release is illustrated in Figure 4A. By incorporating 50, 60 and 65wt.% MPT in the TPUR matrix, the release after 24h is 33, 66 and 100%, respectively. These findings can be correlated to the percolation theory [43-44] as a minimum amount of MPT (percolation threshold) is needed to generate sufficient pores in the inert TPUR matrix in order to ensure sufficient diffusional channels throughout the entire matrix allowing dissolution and release of the entire drug content. Diffusional mass transport (and eventually limited drug solubility effects) can be expected to play an important role in the control of drug release from the TPUR matrices. No changes in tablet dimensions (diameter/height) and tablet geometry (biconvex tablets) were observed for any of the formulations: no swelling or erosion occurred upon wetting of the TPUR matrices. At constant drug load, the drug release profiles were independent of the TPUR grade incorporated in the formulation (data not shown). Figure 4B illustrates the effect of drug solubility on the release profiles (at a 65wt.% drug load). While the highly soluble MPT (aqueous solubility >1000 mg/mL) ensures a complete release after 24h, the

release of Th (10mg/mL) and Dyph (333mg/mL) was, due to their lower aqueous solubility, limited to 20 and 50%, respectively. The addition of a pore former was required to ensure complete release of these drugs: Figure 4C and 4D represent the effect of PEG 4000 (hydrophilic substance) and Tween 80 (surfactant) on the release of Dyph. A gradual increase in drug release is observed in function of the amount of pore-former. The addition of PEG 4000 and Tween 80 to the formulation not only facilitated drug release from the TPUR matrices by generating extra diffusional channels for the API, it also enabled the production of matrices with a higher drug load (>65wt.%) as the melting of PEG 4000 and the liquid Tween 80 phase acted as a lubricant during HME/IM processing. A P539 formulation with a 70% MPT load and 10% PEG 4000 could be processed into high quality tablets; obviously this higher drug load compromised the sustained release capacities of the formulation (complete release already after 12h, data not shown). In addition to the lubricating effect of the pore formers, the addition of 5 and 10% Tween 80 to the formulation (Dyph/P539 65/35) reduced Dyph crystallinity to 76% and 67%, respectively, thus lowering the rotational friction during HMEand improving processability. Similar results were obtained when PEG 4000 was used as pore-former (data not shown). Figure 5 illustrates the correlation between tablet porosity and in-vitro drug release of a MPT/P539 65/35 formulation: the dissolution of interconnecting drug clusters creates additional pores through which the remaining drug can dissolve, creating an empty porous TPUR matrix after 24 h. The formation of additional pores during dissolution and the creation of an empty porous TPUR matrix was confirmed via AFM and SEM experiments (Figure 6). Prior to dissolution, needle-like MPT crystals were detected at the surface of the tablet, whereas after dissolution pores of approximately 10μm were observed.

Raman mapping

To evaluate the distribution homogeneity of the crystalline API, Raman microscopic mapping was performed on the tablets. Six areas were mapped and each area was analyzed using MCR analysis to determine the true underlying factors contributing to the spectral variation. The spectral range studied (800-1500cm^{-1}) contained the fingerprint region of both components of the formulation. No spectral difference was observed in any of the six evaluated area: crystalline API was homogeneously distributed at the surface of the tablet.

Oral toxicity: Simulator human intestinal microbial ecosystem (SHIME)

P523 and T85A (2g/l) were added to the SHIME system to determine its intestinal stability and its possible impact on microbial metabolism indicators (pH, short chain fatty acids [45] and bacteria). No significant differences were observed in bacterial count (Figure 7), short chain fatty acids and pH (data not shown). The polymer structure of T85A was not altered during gastro-intestinal transit as MDSC and FTIR showed similar thermal behavior and spectra, respectively. The structure of P523, on the other hand, was affected as the peaks at 1100, 1275 and 1325cm^{-1} disappeared after exposure to the SHIME media. The first peak was linked to symmetrical ester stretch vibration [46], whereas the other two were assigned to chemical changes in the amorphous SS region of TPUR [46]. Previous research [47-48] already described that polyether-based polyurethanes are more resistant to biodegradation than polyester-based polyurethanes. This first toxicity screening of T85A revealed no evidence of chemical and/or enzymatic reaction after exposure to the intestinal environment, whereas P523 showed signs of ester linkage degradation/hydrolysis. This first indication, regarding TPUR toxicity upon oral administration, was considered positive as the activity or composition of the GI bacterial

community, the pH values and the presence of short chain fatty acids were not altered after exposure to both polymers.

In-vivo evaluation

Figure 8 illustrates the in vitro dissolution profiles and the mean plasma concentration-time profiles after oral administration to beagle dogs of MPT tablets (polyurethane matrix, drug load 65wt.%) and the reference formulation (Slow-Lopresor® 200 Divitabs®, drug load 47wt.%). In vitro dissolution yielded a complete release of MPT after 12 and 24 h for the reference and the polyurethane matrix, respectively. The reference formulation is subjected to surface erosion of its matrix allowing MPT to escape faster due to the increased surface area, while the tablet geometry of the polyurethane matrix remained unchanged after 24 h dissolution experiments. These differences between in vitro drug release patterns were also reflected in their in vivo behavior. Oral administration of the polyurethane formulation resulted in a lower C_{max} and a more sustained release of MPT (up to 16 h) compared to the reference formulation (up to 12 h). These differences in MPT plasma concentration, however, were not statistically significant. Moreover, intact TPUR tablets, which still contained 13% of their initial MPT content, were collected from the faeces of the dogs. No remnants of the reference formulation were found. This is probably attributed to the fast gastro-intestinal (GI) transit time in dogs in combination with a limited amount of fluids in the dog [49], thereby limiting MPT dissolution from the polyurethane matrix. The reference formulation is, compared to the polyurethane matrix, less susceptible to the GI transit time and the limited fluids as surface erosion alters the surface area of the formulation. Other pharmacokinetic parameters (AUC, t_{max} and $HVD_{t50\%Cmax}$) did not differ significantly ($p>0.05$).

Conclusion

This study demonstrated that TPUR polymers are promising matrix formers to produce oral controlled release formulations. Sustained release (in-vitro and in-vivo) of MPT, a highly water-soluble drug, was achieved, while diprophylline required a drug release modifier (Tween 80 or PEG 4000). The high drug load in combination with controlled release capacities is noteworthy in the field of formulations produced via HME/IM.

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